

In the Claims

Please amend the Claims as follows:

1. (Withdrawn) A method for enhancing the solubilisation of at least one proteinaceous macromolecule in a biological sample without inducing substantial acid hydrolysis of said proteinaceous macromolecule, said method comprising incubating the biological sample in a solubilisation reagent at a pH between about pH 1.0 and about pH 6.0.
2. (Withdrawn) The method according to claim 1, wherein the solubilisation reagent has a pH of between about pH 1.0 and about pH 6.0.
3. (Withdrawn) The method according to claim 1, wherein the biological sample is incubated in a solubilisation reagent at a pH of between about pH 2 to about pH 5.
4. (Withdrawn) The method according to claim 1, wherein the biological sample is incubated in a solubilisation reagent at a pH of between about pH 3 to about pH 4.
5. (Withdrawn) The method according to claim 1, wherein the biological sample is incubated in a solubilisation reagent at a pH of between about pH 2 to about pH 3.
6. (Withdrawn) The method according to claim 1, wherein the proteinaceous macromolecule is solubilised in the presence of an aqueous acidic reagent selected from the group consisting of an organic acid solution, inorganic acid solution, acidic buffer, amino acid solution or a mixture thereof
7. (Withdrawn) The method according to claim 6, wherein the organic acid is selected from the group consisting of an ascorbic acid, carboxylic acid and polycarboxylic acid, or a derivative or mixture thereof.
8. (Withdrawn) The method according to claim 7, wherein the carboxylic acid is selected from the group consisting of formic acid, acetic acid, propionic acid, butyric acid, valeric acid, and benzoic acid, or a derivative or mixture thereof.
9. (Withdrawn) The method according to claim 7, wherein the polycarboxylic acid is selected from the group consisting of oxalic acid and citric acid or a derivative or mixture thereof.

10. (Withdrawn) The method according to claim 6, wherein the inorganic acid is selected from the group consisting of phosphoric acid, and orthophosphoric acid or a derivative or mixture thereof.

11. (Withdrawn) The method of claim 6, wherein the acidic buffer comprises acitrophospho buffer.

12. (Withdrawn) The method of claim 1 wherein the solubilisation reagent comprises a chaotropic agent.

13. (Withdrawn) The method of claim 1 wherein the solubilisation reagent comprises a detergent.

14. (Withdrawn) The method according to claim 1 wherein the biological sample is subjected to a physical or chemical means to disrupt the biological sample.

15. (Withdrawn) The method according to claim 1, further comprising recovering the at least one solubilised proteinaceous macromolecule.

16. (Withdrawn) The method according to claim 15, wherein the solubilized proteinaceous macromolecule is recovered by performing a process comprising precipitating the at least one solubilised macromolecule.

17. (Withdrawn) The method according to claim 1, wherein the solubilized proteinaceous macromolecule is recovered by performing a process comprising precipitating and resuspending the protein precipitate.

18. (Withdrawn) The method according to claim 1 further comprising reducing and alkylating the resuspended protein precipitate.

19. (Currently amended) A method of solubilising at least one proteinaceous macromolecule in ~~comprised by~~ a biological sample, without inducing a substantial acid hydrolysis of said proteinaceous macromolecule, said the method comprising:

(i) ~~forming a mixture comprising a buffer, a chaotropic agent and a~~ forming a mixture comprising a buffer, a chaotropic agent and a ~~subjecting the~~ biological sample ~~comprising at least one proteinaceous macromolecule,~~ comprising at least one proteinaceous macromolecule, ~~to a physical or chemical means to disrupt said biological sample and incubating the biological sample in the~~

presence of a reagent at a pH between about pH 1.0 and pH 7, to thereby solubilize at least one proteinaceous macromolecule in the biological sample; and

(ii) disrupting the biological sample in the presence of the buffer and the chaotropic agent, at a pH between about pH 1.0 and about pH 6.0, performing one or more processes selected from the group consisting of:

(a) recovering the solubilized proteinaceous macromolecule by performing a process comprising precipitating one or more proteins in the extract at (i) to thereby precipitate at least the solubilized proteinaceous macromolecule and resuspending the precipitated proteinaceous macromolecule;

(b) reducing and alkylating the solubilized proteinaceous macromolecule at (i) or the resuspended proteinaceous macromolecule at (ii)(a); and

(c) subjecting the solubilized proteinaceous macromolecule at (i) or the resuspended proteinaceous macromolecule at (ii)(a) or the reduced and alkylated proteinaceous macromolecule at (ii)(b) to a resolving means for a time and under conditions sufficient to resolve the proteinaceous macromolecule from other macromolecules present in the biological sample and then identifying the resolved proteinaceous macromolecule.

20-23 (Canceled).

24. (Withdrawn) A kit for enhancing solubilisation of a proteinaceous macromolecule a biological sample without inducing substantial acid hydrolysis of said proteinaceous macromolecule, the kit comprising a solubilisation reagent to solubilise at least one macromolecule in a biological sample, wherein the solubilisation reagent has a pH of about pH 1 to about pH 6 and optionally comprising directions to solubilise and/or recover a macromolecule in a biological sample, and/or directions to resolve a macromolecule in a biological sample.

25. (Withdrawn) A proteinaceous macromolecule solubilised by the method according to claim 1.

26. (Withdrawn) Use of an acidic reagent having a pH of about pH 1 to about pH 6 in the preparation of an solubilisation reagent solution for use in solubilising a proteinaceous

macromolecule from a biological sample, without inducing substantial acid hydrolysis of said proteinaceous macromolecule.

27. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 19, wherein the disrupting the biological sample is selected from the group consisting of enzymatically lysing the sample, grinding the sample, cooling the sample in liquid nitrogen, glass bead beating the sample, incubating the sample in toluene, sonicating the sample, and a combination thereof.

28. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 19, wherein the disrupting the biological sample comprises sonicating the sample.

29. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 19, wherein the pH is between about pH 2 and pH 5.

30. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 19, wherein the buffer comprises an acid selected from the group consisting of ascorbic acid, benzoic acid, formic acid, acetic acid, propionic acid, butyric acid, valeric acid, an amino acid, citric acid, ascorbic acid, orthophosphoric acid and a combination thereof.

31. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 19, wherein the chaotropic agent is selected from the group consisting of urea, thiourea, and a mixture thereof.

32. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 19, further comprising adding a detergent to the mixture.

33. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 32, wherein the detergent is selected from the group consisting of 3-(4-heptyl) phenyl 3-hydroxypropyl dimethyl ammonio propane sulfonate (C7Bz0) and tetradecanoylamido propyl dimethyl ammonio propane sulfonate (ASB 14).

34. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 19, further comprising reducing the at least one proteinaceous macromolecule, wherein the reducing comprises contacting the at least one proteinaceous macromolecule with a reducing agent selected from the group consisting of dithiothreitol (DTT), tri-n-butylphosphine (TBP), beta-mercaptoethanol, iodoacetamide, and a mixture thereof.

35. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 36, further comprising alkylating the at least one proteinaceous macromolecule, wherein the alkylating comprises contacting the at least one proteinaceous macromolecule with an alkylating agent selected from the group consisting of iodoacetamide, vinyl pyridine, acrylamide, iodoacetic acid and a mixture thereof.

36. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with claim 19, wherein the solubilising occurs without substantial acid-induced hydrolysis of the at least one proteinaceous macromolecule.

37. (New) A method of forming an extract of a biological sample comprising at least one proteinaceous macromolecule, the method comprising:

solubilising at least one proteinaceous macromolecule comprised by a biological sample in accordance with the method of claim 19; and
precipitating the at least one proteinaceous macromolecule to form a precipitate.

38. (New) A method of forming an extract in accordance with claim 37 wherein the precipitating comprises adding to the mixture a precipitating agent selected from the group consisting of ammonium sulfate, polyethylene glycol (PEG) and an organic solvent selected from the group consisting of methanol, acetone and a mixture thereof.

39. (New) A method of forming an extract in accordance with claim 38, further comprising resuspending the precipitate.

40. (New) A method of forming an extract in accordance with claim 39, wherein the resuspending the precipitate comprises resuspending the precipitate in an extraction solution, wherein the extraction solution is selected from the group consisting of an organic acid buffer, an inorganic acid buffer, an amino acid solution, and a mixture thereof.

41. (New) A method of forming an extract in accordance with claim 40, wherein the extraction solution is an acidic aqueous solvent having a pH of about pH 1.0 to about pH 6.0.

42. (New) A method of forming an extract in accordance with claim 40, wherein the extraction solution has a pH of about 7.0.

43. (New) A method of forming an extract in accordance with claim 40, wherein the extraction solution has an alkaline pH.

44. (New) A method of forming an extract in accordance with claim 40, wherein the extraction solution has a pH of 10.4.

45. (New) A method of forming an extract in accordance with claim 39, further comprising reducing the at least one proteinaceous macromolecule, wherein the reducing comprises contacting the at least one proteinaceous macromolecule with a reducing agent selected from the group consisting of dithiothreitol (DTT), tri-n-butylphosphine (TBP), beta-mercaptoethanol, iodoacetamide, and a mixture thereof.

46. (New) A method of forming an extract in accordance with claim 45, further comprising alkylating the at least one proteinaceous macromolecule, wherein the alkylating comprises contacting the at least one proteinaceous macromolecule with an alkylating agent selected from the group consisting of iodoacetamide, vinyl pyridine, acrylamide, iodoacetic acid and a mixture thereof.

47. (New) A method of forming an extract in accordance with claim 19, wherein the disrupting comprises incubating the mixture.

48. (New) A method of solubilising at least one proteinaceous macromolecule comprised by a biological sample, the method comprising:

(i) forming a mixture comprising a biological sample comprising at least one proteinaceous macromolecule, a buffer and a detergent selected from the group consisting of a cationic detergent, an anionic detergent and a non-ionic detergent., at a pH between about pH 1.0 and pH 7; and

(ii) disrupting the biological sample in the presence of the buffer and the detergent, at a pH between about pH 1.0 and pH 7.

49. (New) A method in accordance with claim 48, wherein the pH is between about pH 2 to about pH 5.

50. (New) A method in accordance with claim 48, wherein the detergent is selected from the group consisting of 3-(4-heptyl) phenyl 3-hydroxypropyl dimethyl ammonio propane sulfonate (C7Bz0), and tetradecanoylamido propyl dimethyl ammonio propane sulfonate (ASB 14).

51. (New) A method in accordance with claim 48, wherein the mixture further comprises a chaotropic agent.